

## CLAIMS

We claim:

1. A lyophilization medium for a microorganism wherein the medium is substantially free of animal-derived products and comprises yeast extract and monosodium glutamate.
2. The lyophilization medium of claim 1, comprising about 1-10% (w/v) monosodium glutamate and about 1-10% (w/v) yeast extract.
3. The lyophilization medium of claim 2, comprising about 5 % (w/v) monosodium glutamate and about 10% (w/v) yeast extract.
4. The lyophilization medium of claim 1 or 2 or 3 wherein the microorganism is a strain of bacteria.
5. The lyophilization medium of claim 4 wherein the strain of bacteria is *Corynebacterium diphtheriae*
6. A method for preparing a freeze-dried culture of a microorganism comprising the steps of:  
providing a quantity of the microorganism;  
mixing said quantity with a lyophilization medium wherein the medium is substantially free of animal-derived products and comprises yeast extract and monosodium glutamate to provide a mixture; and  
freeze-drying said mixture.
7. The method of claim 4, wherein the lyophilization medium of comprises about 5 % (w/v) monosodium glutamate and about 10% (w/v) yeast extract.

8. The method of claim 5, wherein the lyophilization medium of comprises about 1-10% (w/v) monosodium glutamate and about 1-10% (w/v) yeast extract.
9. The method of claim 6 or 7 or 8 wherein freeze-drying of said mixture comprises steps of:
  - (a) achieving a first temperature of about  $-30^{\circ}\text{C}$  for said mixture to provide a cooled mixture;
  - (b) maintaining said cooled mixture in a vacuum for a time until said cooled mixture is substantially dry to provide a dried mixture.
10. The method of claim 7 wherein the vacuum is about 120 mT.
11. The method of claim 8 wherein the time is between about 10 and about 12 hours.
12. The method of claim 7 wherein the step of maintaining said cooled mixture in a vacuum for a time until said cooled mixture is substantially dry to provide a dried mixture comprises:
  - (a) maintaining said cooled mixture in a vacuum for a time of between about 10 and about 12 hours; and
  - (b) increasing said temperature of about  $-30^{\circ}\text{C}$  to a second temperature of about  $+20^{\circ}\text{C}$ .
13. The method of claim 10 wherein the vacuum is about 120 mT.
14. The method of claim 6 or 7 or 8 wherein the microorganism is a strain of bacteria.
15. The method of claim 14 wherein the strain of bacteria is a strain of *Corynebacterium diphtheriae*

16. A free-dried lyophile comprising cells of a microorganism and a lyophilization medium wherein the medium is substantially free of animal-derived products and comprises yeast extract and monosodium glutamate.
17. The freeze-dried lyophile of claim 12, wherein the medium comprises about 1-10% (w/v) monosodium glutamate and about 1-10% (w/v) yeast extract.
18. The freeze-dried lyophile of claim 13, wherein the medium comprises about 5 % (w/v) monosodium glutamate and about 10% (w/v) yeast extract.
19. The freeze-dried lyophile of claim 16 or 17 or 18 wherein the microorganism is a strain of bacteria.
20. The freeze-dried lyophile of claim 19 wherein the strain of bacteria is a strain of *Corynebacterium diphtheriae*.

Figure 1: A flow diagram outlining the preparation and lyophilization of a *C. diphtheriae* culture.